

WHAT IS CLAIMED IS:

1. An isolated nucleic acid molecule comprising:
 - (a) one or more recombination sites; and
 - (b) one or more nucleic acid sequences which encode an amino acid sequence tag.
2. The isolated nucleic acid molecule of claim 1, further comprising at least one additional nucleic acid sequence selected from the group consisting of a selectable marker, a cloning site, a restriction site, a promoter, an operator, an operon, a nucleotide sequence encoding a gene product which allows for negative selection, an origin of replication, a nucleotide sequence which encodes a repressor of at least one promoter, and a gene or partial gene.
3. The isolated nucleic acid molecule of claim 1, wherein a nucleic acid sequence of interest can be inserted at or within 20 nucleotides of said one or more recombination sites, thereby producing a polynucleotide construct that encodes a fusion protein, said fusion protein comprising: (i) said amino acid sequence tag; and (ii) the amino acid sequence encoded by said nucleic acid sequence of interest.
4. The isolated nucleic acid molecule of claim 1, further comprising a nucleic acid sequence that encodes an amino acid sequence that is capable of being cleaved by one or more proteases.
5. The isolated nucleic acid molecule of claim 4, wherein said amino acid sequence that is capable of being cleaved by one or more proteases is an amino acid sequence that is capable of being cleaved by enterokinase.
6. The isolated nucleic acid molecule of claim 4, wherein a nucleic acid sequence of interest can be inserted at or within 20 nucleotides of said one

or more recombination sites thereby producing a polynucleotide construct that encodes a fusion protein, said fusion protein comprising: (i) said amino acid sequence that is capable of being cleaved by one or more proteases, flanked on one side by (ii) said amino acid tag, and on the other side by (iii) the amino acid sequence encoded by said nucleic acid sequence of interest.

7. The nucleic acid molecule of claim 1, wherein said amino acid sequence tag is an amino acid sequence that is capable of being post-translationally modified.
8. The isolated nucleic acid molecule of claim 7, wherein said amino acid sequence that is capable of being post-translationally modified is an amino acid sequence that is capable of being post-translationally modified by biotinylation, attachment of 4-phosphopanthetheine, attachment of lipoic acid or attachment of flavins.
9. The isolated nucleic acid molecule of claim 7, wherein said amino acid sequence that is capable of being post-translationally modified is an amino acid sequence that is capable of being biotinylated.
10. The isolated nucleic acid molecule of claim 9, wherein said amino acid sequence that is capable of being biotinylated is all or a portion of the *Klebsiella pneumoniae* oxalacetate decarboxylase α subunit, all or a portion of the *Propionibacterium shermanii* transcarboxylase 1.3S subunit, or all or a portion of the *Escherichia coli* biotin carboxyl carrier protein component of acetyl-CoA carboxylase.
11. The isolated nucleic acid molecule of claim 9, wherein said amino acid sequence that is capable of being biotinylated is a portion of the C-

terminus of the *Klebsiella pneumoniae* oxalacetate decarboxylase α subunit.

12. The isolated nucleic acid molecule of claim 11, wherein said amino acid sequence that is capable of being biotinylated is the BIOTAG™.
13. The isolated nucleic acid molecule of claim 1, wherein said nucleic acid molecule is a circular molecule.
14. The isolated nucleic acid molecule of claim 1, wherein said nucleic acid molecule comprises two or more recombination sites.
15. The isolated nucleic acid molecule of claim 1, wherein said recombination sites are selected from the group consisting of: (a) *attB* sites, (b) *attP* sites, (c) *attL* sites, (d) *attR* sites, (e) *lox* sites, (f) *psi* sites, (g) *dif* sites, (h) *cer* sites, (i) *frt* sites, and mutants, variants, and derivatives of the recombination sites of (a), (b), (c), (d), (e), (f), (g), (h), or (i) which retain the ability to undergo recombination.
16. A vector comprising the isolated nucleic acid molecule of claim 1.
17. A host cell comprising the isolated nucleic acid molecule of claim 1.
18. A host cell comprising the vector of claim 16.
19. A method of producing a polynucleotide construct that encodes a fusion protein that comprises an amino acid sequence tag, said method comprising:
 - (a) obtaining a first nucleic acid molecule comprising a nucleotide sequence of interest flanked by at least a first and at least a second recombination sites that do not recombine with each other;

- (b) obtaining a second nucleic acid molecule comprising: (i) at least a third and fourth recombination sites that do not recombine with each other; and (ii) one or more nucleic acid sequences which encode an amino acid sequence tag; and
 - (c) contacting said first nucleic acid molecule with said second nucleic acid molecule under conditions favoring recombination between said first and third and between said second and fourth recombination sites, thereby producing a product polynucleotide construct;
wherein said product polynucleotide construct encodes a fusion protein comprising: (i) said amino acid sequence tag; and (ii) the amino acid sequence encoded by said nucleotide acid sequence of interest.
20. The method of claim 19, wherein said second nucleic acid molecule further comprises a nucleic acid sequence that encodes an amino acid sequence that is capable of being cleaved by one or more proteases; and
wherein said product polynucleotide construct encodes a fusion protein comprising: (i) said amino acid sequence that is capable of being cleaved by one or more proteases, flanked on one side by (ii) said amino acid sequence tag, and on the other side by (iii) the amino acid sequence encoded by said nucleotide sequence of interest.
21. The method of claim 20, wherein said amino acid sequence that is capable of being cleaved by one or more proteases is an amino acid sequence that is capable of being cleaved by enterokinase.
22. The method of claim 19, wherein said amino acid sequence tag is an amino acid sequence that is capable of being post-translationally modified.
23. The method of claim 22, wherein said amino acid sequence that is capable of being post-translationally modified is an amino acid sequence that is capable of being post-translationally modified by biotinylation, attachment

of 4-phosphopanthetheine, attachment of lipoic acid or attachment of flavins.

24. The method of claim 22, wherein said amino acid sequence that is capable of being post-translationally modified is an amino acid sequence that is capable of being biotinylated.
25. The method of claim of claim 24, wherein said amino acid sequence that is capable of being biotinylated is all or a portion of the *Klebsiella pneumoniae* oxalacetate decarboxylase α subunit, all or a portion of the *Propionibacterium shermanii* transcarboxylase 1.3S subunit, or all or a portion of the *Escherichia coli* biotin carboxyl carrier protein component of acetyl-CoA carboxylase.
26. The method of claim of claim 24, wherein said amino acid sequence that is capable of being biotinylated is a portion of the C-terminus of the *Klebsiella pneumoniae* oxalacetate decarboxylase α subunit.
27. The method of claim 26, wherein said amino acid sequence that is capable of being biotinylated is the BIOTAG™.
28. The method of claim 19, wherein said second nucleic acid molecule is a vector.
29. The method of claim 19, wherein said first nucleic acid molecule is a circular nucleic acid molecule.
30. The method of claim 19, wherein said first nucleic acid molecule is a linear nucleic acid molecule.

31. The method of claim 30, wherein said first nucleic acid molecule is a PCR product.
32. The method of claim 19, further comprising inserting said product polynucleotide construct into a host cell.
33. The method of claim 20, further comprising inserting said product polynucleotide construct into a host cell.
34. The method of claim 19, wherein said second nucleic acid molecule comprises at least one additional nucleic acid sequence selected from the group consisting of a selectable marker, a cloning site, a restriction site, a promoter, an operator, an operon, a nucleotide sequence encoding a gene product which allows for negative selection, an origin of replication, a nucleotide sequence which encodes a repressor of at least one promoter, and a gene or partial gene.
35. The method of claim 19, wherein said first, second, third and fourth recombination sites are selected from the group consisting of: (a) *attB* sites, (b) *attP* sites, (c) *attL* sites, (d) *attR* sites, (e) *lox* sites, (f) *psi* sites, (g) *dif* sites, (h) *cer* sites, (i) *frt* sites, and mutants, variants, and derivatives of the recombination sites of (a), (b), (c), (d), (e), (f), (g), (h), or (i) which retain the ability to undergo recombination.
36. The method of claim 19, wherein said first and said second nucleic acid molecules are combined in the presence of at least one recombination protein.
37. The method of claim 36, wherein said recombination protein is selected from the group consisting of: (a) Cre, (b) Int, (c) IHF, (d) Xis, (e) Fis, (f) Hin, (g) Gin, (h) Cin, (i) Tn3 resolvase, (j) TndX, (k) XerC, and (l) XerD.

38. The method of claim 36, wherein said recombination protein is Cre.
39. An isolated nucleic acid molecule comprising:
- (a) one or more topoisomerase recognition sites and/or one or more topoisomerases; and
 - (b) one or more nucleic acid sequences which encode an amino acid sequence tag.
40. The isolated nucleic acid molecule of claim 39, further comprising at least one additional nucleic acid sequence selected from the group consisting of a selectable marker, a cloning site, a restriction site, a promoter, an operator, an operon, a nucleotide sequence encoding a gene product which allows for negative selection, an origin of replication, a nucleotide sequence which encodes a repressor of at least one promoter, and a gene or partial gene.
41. The isolated nucleic acid molecule of claim 39, wherein a nucleic acid sequence of interest can be inserted at or within 20 nucleotides of said one or more topoisomerase recognition sites and/or at or within 20 nucleotide of the position of said one or more topoisomerases, thereby producing a polynucleotide construct that encodes a fusion protein, said fusion protein comprising: (i) said amino acid sequence tag; and (ii) the amino acid sequence encoded by said nucleic acid sequence of interest.
42. The isolated nucleic acid molecule of claim 39, further comprising a nucleic acid sequence that encodes an amino acid sequence that is capable of being cleaved by one or more proteases.
43. The isolated nucleic acid molecule of claim 42, wherein said amino acid sequence that is capable of being cleaved by one or more proteases is an amino acid sequence that is capable of being cleaved by enterokinase.

44. The isolated nucleic acid molecule of claim 42, wherein a nucleic acid sequence of interest can be inserted at or within 20 nucleotides of said one or more topoisomerase recognition sites and/or at the position of said one or more topoisomerases thereby producing a polynucleotide construct that encodes a fusion protein, said fusion protein comprising: (i) said amino acid sequence that is capable of being cleaved by one or more proteases, flanked on one side by (ii) said amino acid tag, and on the other side by (iii) the amino acid sequence encoded by said nucleic acid sequence of interest.
45. The isolated nucleic acid molecule of claim 39, wherein said amino acid sequence tag is an amino acid sequence that is capable of being post-translationally modified.
46. The isolated nucleic acid molecule of claim 45, wherein said amino acid sequence that is capable of being post-translationally modified is an amino acid sequence that is capable of being post-translationally modified by biotinylation, attachment of 4-phosphopanthetheine, attachment of lipoic acid or attachment of flavins.
47. The isolated nucleic acid molecule of claim 45, wherein said amino acid sequence that is capable of being post-translationally modified is an amino acid sequence that is capable of being biotinylated.
48. The isolated nucleic acid molecule of claim 47, wherein said amino acid sequence that is capable of being biotinylated is all or a portion of the *Klebsiella pneumoniae* oxalacetate decarboxylase α subunit, all or a portion of the *Propionibacterium shermanii* transcarboxylase 1.3S subunit, or all or a portion of the *Escherichia coli* biotin carboxyl carrier protein component of acetyl-CoA carboxylase.

49. The isolated nucleic acid molecule of claim 47, wherein said amino acid sequence that is capable of being biotinylated is a portion of the C-terminus of the *Klebsiella pneumoniae* oxalacetate decarboxylase α subunit.
50. The isolated nucleic acid molecule of claim 49, wherein said amino acid sequence that is capable of being biotinylated is the BIOTAG™.
51. The isolated nucleic acid molecule of claim 39, wherein said nucleic acid molecule is a circular molecule.
52. The isolated nucleic acid molecule of claim 39, wherein said nucleic acid molecule comprises two or more recombination sites.
53. The isolated nucleic acid molecule of claim 39, wherein said topoisomerase is a type I topoisomerase.
54. The isolated nucleic acid molecule of claim 53, wherein said type I topoisomerase is a type IB topoisomerase.
55. The isolated nucleic acid molecule of claim 54, wherein said type IB topoisomerase is selected from the group consisting of eukaryotic nuclear type I topoisomerase and a poxvirus topoisomerase.
56. The isolated nucleic acid molecule of claim 55, wherein said poxvirus topoisomerase is produced by or isolated from a virus selected from the group consisting of vaccinia virus, Shope fibroma virus, ORF virus, fowlpox virus, molluscum contagiosum virus and *Amsacta moorei* entomopoxvirus.
57. A vector comprising the isolated nucleic acid molecule of claim 39.

58. A host cell comprising the isolated nucleic acid molecule of claim 39.

59. A host cell comprising the vector of claim 57.

60. A method of producing a polynucleotide construct that encodes a fusion protein that comprises an amino acid sequence tag, said method comprising:

- (a) obtaining a first nucleic acid molecule comprising a nucleotide sequence of interest;
- (b) obtaining a second nucleic acid molecule comprising at least two topoisomerase recognition sites, at least one topoisomerase, and at least one nucleic acid sequence which encodes an amino acid sequence tag;
- (c) mixing said first nucleic acid molecule with said second nucleic acid molecule; and
- (d) incubating said mixture under conditions such that said first nucleic acid molecule is inserted into said second nucleic acid molecule between said at least two topoisomerase recognition sites, thereby producing a product polynucleotide construct;

wherein said product polynucleotide construct encodes a fusion protein comprising: (i) said amino acid sequence tag; and (ii) the amino acid sequence encoded by said nucleotide sequence of interest.

61. The method of claim 60, wherein said second nucleic acid molecule further comprises a nucleic acid sequence that encodes an amino acid sequence that is capable of being cleaved by one or more proteases; and

wherein said product polynucleotide construct encodes a fusion protein comprising: (i) said amino acid sequence that is capable of being cleaved by one or more proteases, flanked on one side by (ii) said amino acid sequence tag, and on the other side by (iii) the amino acid sequence encoded by said nucleotide sequence of interest.

62. The method of claim 61, wherein said amino acid sequence that is capable of being cleaved by one or more proteases is an amino acid sequence that is capable of being cleaved by enterokinase.
63. The method of claim 60, wherein said amino acid sequence tag is an amino acid sequence that is capable of being post-translationally modified.
64. The method of claim 63, wherein said amino acid sequence that is capable of being post-translationally modified is an amino acid sequence that is capable of being post-translationally modified by biotinylation, attachment of 4-phosphopanthetheine, attachment of lipoic acid or attachment of flavins.
65. The method of claim 63, wherein said amino acid sequence that is capable of being post-translationally modified is an amino acid sequence that is capable of being biotinylated.
66. The method of claim of claim 65, wherein said amino acid sequence that is capable of being biotinylated is all or a portion of the *Klebsiella pneumoniae* oxalacetate decarboxylase α subunit, all or a portion of the *Propionibacterium shermanii* transcarboxylase 1.3S subunit, or all or a portion of the *Escherichia coli* biotin carboxyl carrier protein component of acetyl-CoA carboxylase.
67. The method of claim of claim 65, wherein said amino acid sequence that is capable of being biotinylated is a portion of the C-terminus of the *Klebsiella pneumoniae* oxalacetate decarboxylase α subunit.
68. The method of claim 67, wherein said amino acid sequence that is capable of being biotinylated is the BIOTAG™.

69. The method of claim 60, wherein said second nucleic acid molecule is a vector.
70. The method of claim 60, wherein said first nucleic acid molecule is a linear nucleic acid molecule.
71. The method of claim 70, wherein said first nucleic acid molecule is a blunt-end nucleic acid molecule.
72. The method of claim 60, wherein said first nucleic acid molecule is a PCR product.
73. The method of claim 60, further comprising inserting said product polynucleotide construct into a host cell.
74. The method of claim 61, further comprising inserting said product polynucleotide construct into a host cell.
75. The method of claim 60, wherein said second nucleic acid molecule comprises at least one additional nucleic acid sequence selected from the group consisting of a selectable marker, a cloning site, a restriction site, a promoter, an operator, an operon, a nucleotide sequence encoding a gene product which allows for negative selection, an origin of replication, a nucleotide sequence which encodes a repressor of at least one promoter, and a gene or partial gene.
76. The method of claim 60, wherein said topoisomerase is a type I topoisomerase.
77. The method of claim 76, wherein said type I topoisomerase is a type IB topoisomerase.

78. The method of claim 77, wherein said type IB topoisomerase is selected from the group consisting of eukaryotic nuclear type I topoisomerase and a poxvirus topoisomerase.
79. The method of claim 78, wherein said poxvirus topoisomerase is produced by or isolated from a virus selected from the group consisting of vaccinia virus, Shope fibroma virus, ORF virus, fowlpox virus, molluscum contagiosum virus and *Amsacta moorei* entomopoxvirus.
80. An isolated nucleic acid molecule comprising:
- (a) one or more recombination sites;
 - (b) one or more topoisomerase recognition sites and/or one or more topoisomerases; and
 - (c) one or more nucleic acid sequences which encode an amino acid sequence tag.
81. The isolated nucleic acid molecule of claim 80, further comprising at least one additional nucleic acid sequence selected from the group consisting of a selectable marker, a cloning site, a restriction site, a promoter, an operator, an operon, a nucleotide sequence encoding a gene product which allows for negative selection, an origin of replication, a nucleotide sequence which encodes a repressor of at least one promoter, and a gene or partial gene.
82. The isolated nucleic acid molecule of claim 80, wherein a nucleic acid sequence of interest can be inserted at or within 20 nucleotides of said one or more recombination sites, thereby producing a polynucleotide construct that encodes a fusion protein, said fusion protein comprising: (i) said amino acid sequence tag; and (ii) the amino acid sequence encoded by said nucleic acid sequence of interest.

83. The isolated nucleic acid molecule of claim 80, wherein a nucleic acid sequence of interest can be inserted at or within 20 nucleotides of said one or more topoisomerase recognition sites and/or at or within 20 nucleotides of the position of said one or more topoisomerases, thereby producing a polynucleotide construct that encodes a fusion protein, said fusion protein comprising: (i) said amino acid tag; and (ii) the amino acid sequence encoded by said nucleic acid sequence of interest.
84. The isolated nucleic acid molecule of claim 80, further comprising a nucleic acid sequence that encodes an amino acid sequence that is capable of being cleaved by one or more proteases.
85. The isolated nucleic acid molecule of claim 84, wherein said amino acid sequence that is capable of being cleaved by one or more proteases is an amino acid sequence that is capable of being cleaved by enterokinase.
86. The isolated nucleic acid molecule of claim 84, wherein a nucleic acid sequence of interest can be inserted at or within 20 nucleotides of said one or more recombination sites, thereby producing a polynucleotide construct that encodes a fusion protein, said fusion protein comprising: (i) said amino acid sequence that is capable of being cleaved by one or more proteases, flanked on one side by (ii) said amino acid sequence tag, and on the other side by (iii) the amino acid sequence encoded by said nucleic acid sequence of interest.
87. The isolated nucleic acid molecule of claim 84, wherein a nucleic acid sequence of interest can be inserted at or within 20 nucleotides of said one or more topoisomerase recognition sites and/or at or within 20 nucleotides of the position of said one or more topoisomerases, thereby producing a polynucleotide construct that encodes a fusion protein, said fusion protein comprising: (i) said amino acid sequence that is capable of being cleaved by one or more proteases, flanked on one side by (ii) said amino acid

sequence tag, and on the other side by (iii) the amino acid sequence encoded by said nucleic acid sequence of interest.

88. The isolated nucleic acid molecule of claim 80, wherein said amino acid sequence tag is an amino acid sequence that is capable of being post-translationally modified.

89. The isolated nucleic acid molecule of claim 88, wherein said amino acid sequence that is capable of being post-translationally modified is an amino acid sequence that is capable of being post-translationally modified by biotinylation, attachment of 4-phosphopanthetheine, attachment of lipoic acid or attachment of flavins.

90. The isolated nucleic acid molecule of claim 80, wherein said amino acid sequence that is capable of being post-translationally modified is an amino acid sequence that is capable of being biotinylated.

91. The isolated nucleic acid molecule of claim 90, wherein said amino acid sequence that is capable of being biotinylated is all or a portion of the *Klebsiella pneumoniae* oxalacetate decarboxylase α subunit, all or a portion of the *Propionibacterium shermanii* transcarboxylase 1.3S subunit, or all or a portion of the *Escherichia coli* biotin carboxyl carrier protein component of acetyl-CoA carboxylase.

92. The isolated nucleic acid molecule of claim 90, wherein said amino acid sequence that is capable of being biotinylated is a portion of the C-terminus of the *Klebsiella pneumoniae* oxalacetate decarboxylase α subunit.

93. The isolated nucleic acid molecule of claim 92, wherein said amino acid sequence that is capable of being biotinylated is the BIOTAG™.

94. The isolated nucleic acid molecule of claim 80, wherein said nucleic acid molecule is a circular molecule.
95. The isolated nucleic acid molecule of claim 80, wherein said nucleic acid molecule comprises two or more recombination sites.
96. The isolated nucleic acid molecule of claim 80, wherein said recombination sites are selected from the group consisting of: (a) *attB* sites, (b) *attP* sites, (c) *attL* sites, (d) *attR* sites, (e) *lox* sites, (f) *psi* sites, (g) *dif* sites, (h) *cer* sites, (i) *frt* sites, and mutants, variants, and derivatives of the recombination sites of (a), (b), (c), (d), (e), (f), (g), (h), or (i) which retain the ability to undergo recombination.
97. The isolated nucleic acid molecule of claim 80, wherein said topoisomerase is a type I topoisomerase.
98. The isolated nucleic acid molecule of claim 97, wherein said type I topoisomerase is a type IB topoisomerase.
99. The isolated nucleic acid molecule of claim 98, wherein said type IB topoisomerase is selected from the group consisting of eukaryotic nuclear type I topoisomerase and a poxvirus topoisomerase.
100. The isolated nucleic acid molecule of claim 99, wherein said poxvirus topoisomerase is produced by or isolated from a virus selected from the group consisting of vaccinia virus, Shope fibroma virus, ORF virus, fowlpox virus, molluscum contagiosum virus and *Amsacta moorei* entomopoxvirus.
101. A vector comprising the isolated nucleic acid molecule of claim 80.

102. A host cell comprising the isolated nucleic acid molecule of claim 80.
103. A host cell comprising the vector of claim 101.
104. A method of producing a polynucleotide construct that encodes a fusion protein that comprises an amino acid sequence tag, said method comprising:
 - (a) obtaining a first nucleic acid molecule comprising a nucleotide sequence of interest;
 - (b) obtaining a second nucleic acid molecule comprising (i) at least a first topoisomerase recognition site flanked by (ii) at least a first recombination site, and (iii) at least a second topoisomerase recognition site flanked by (iv) at least a second recombination site, wherein said first and second recombination sites do not recombine with each other, and (v) at least one topoisomerase;
 - (c) obtaining a third nucleic acid molecule comprising: (i) at least a third and fourth recombination sites that do not recombine with each other; and (ii) one or more nucleic acid sequences which encode an amino acid sequence tag;
 - (d) mixing said first nucleic acid molecule with said second nucleic acid molecule;
 - (e) incubating said mixture under conditions such that said first nucleic acid molecule is inserted into said second nucleic acid molecule between said at least two topoisomerase recognition sites, thereby producing a first product polynucleotide construct;
 - (f) contacting said first product polynucleotide construct with said third nucleic acid molecule under conditions favoring recombination between said first and third and between said second and fourth recombination sites, thereby producing a second product polynucleotide construct;

wherein said second product polynucleotide construct encodes a fusion protein comprising: (i) said amino acid sequence tag; and (ii) the amino acid sequence encoded by said nucleotide sequence of interest.

105. The method of claim 104, wherein said third nucleic acid molecule further comprises a nucleic acid sequence that encodes an amino acid sequence that is capable of being cleaved by one or more proteases; and wherein said second product polynucleotide construct encodes a fusion protein comprising: (i) said amino acid sequence that is capable of being cleaved by one or more proteases, flanked on one side by (ii) said amino acid sequence tag, and on the other side by (iii) the amino acid sequence encoded by said nucleotide sequence of interest.

106. The method of claim 105, wherein said amino acid sequence that is capable of being cleaved by one or more proteases is an amino acid sequence that is capable of being cleaved by enterokinase.

107. The method of claim 104, wherein said amino acid sequence tag is an amino acid sequence that is capable of being post-translationally modified.

108. The method of claim 107, wherein said amino acid sequence that is capable of being post-translationally modified is an amino acid sequence that is capable of being post-translationally modified by biotinylation, attachment of 4-phosphopantetheine, attachment of lipoic acid or attachment of flavins.

109. The method of claim 107, wherein said amino acid sequence that is capable of being post-translationally modified is an amino acid sequence that is capable of being biotinylated.

110. The method of claim of claim 109, wherein said amino acid sequence that is capable of being biotinylated is all or a portion of the *Klebsiella*

pneumoniae oxalacetate decarboxylase α subunit, all or a portion of the *Propionibacterium shermanii* transcarboxylase 1.3S subunit, or all or a portion of the *Escherichia coli* biotin carboxyl carrier protein component of acetyl-CoA carboxylase.

111. The method of claim of claim 109, wherein said amino acid sequence that is capable of being biotinylated is a portion of the C-terminus of the *Klebsiella pneumoniae* oxalacetate decarboxylase α subunit.
112. The method of claim 111, wherein said amino acid sequence that is capable of being biotinylated is the BIOTAG™.
113. The method of claim 104, wherein said second nucleic acid molecule is a vector.
114. The method of claim 104, wherein said third nucleic acid molecule is a vector.
115. The method of claim 104, wherein said first nucleic acid molecule is a linear nucleic acid molecule.
116. The method of claim 115, wherein said first nucleic acid molecule is a blunt-end nucleic acid molecule.
117. The method of claim 104, wherein said first nucleic acid molecule is a PCR product.
118. The method of claim 104, further comprising inserting said first product polynucleotide construct into a host cell.

119. The method of claim 104, further comprising inserting said second product polynucleotide construct into a host cell.
120. The method of claim 104, wherein said second and/or said third nucleic acid molecules comprises at least one additional nucleic acid sequence selected from the group consisting of a selectable marker, a cloning site, a restriction site, a promoter, an operator, an operon, a nucleotide sequence encoding a gene product which allows for negative selection, an origin of replication, a nucleotide sequence which encodes a repressor of at least one promoter, and a gene or partial gene.
121. The method of claim 104, wherein said first, second, third and fourth recombination sites are selected from the group consisting of: (a) *attB* sites, (b) *attP* sites, (c) *attL* sites, (d) *attR* sites, (e) *lox* sites, (f) *psi* sites, (g) *dif* sites, (h) *cer* sites, (i) *frt* sites, and mutants, variants, and derivatives of the recombination sites of (a), (b), (c), (d), (e), (f), (g), (h), or (i) which retain the ability to undergo recombination.
122. The method of claim 104, wherein said topoisomerase is a type I topoisomerase.
123. The method of claim 122, wherein said type I topoisomerase is a type IB topoisomerase.
124. The method of claim 123, wherein said type IB topoisomerase is selected from the group consisting of eukaryotic nuclear type I topoisomerase and a poxvirus topoisomerase.
125. The method of claim 124, wherein said poxvirus topoisomerase is produced by or isolated from a virus selected from the group consisting of vaccinia virus, Shope fibroma virus, ORF virus, fowlpox virus, molluscum contagiosum virus and *Amsacta moorei* entomopoxvirus.

126. The method of claim 104, wherein said first product polynucleotide construct and said third nucleic acid molecule are combined in the presence of at least one recombination protein.
127. The method of claim 126, wherein said recombination protein is selected from the group consisting of: (a) Cre, (b) Int, (c) IHF, (d) Xis, (e) Fis, (f) Hin, (g) Gin, (h) Cin, (i) Tn3 resolvase, (j) TndX, (k) XerC, and (l) XerD.
128. The method of claim 126, wherein said recombination protein is Cre.
129. A vector selected from the group consisting of pET104-DEST, pET104/GW/*lacZ*, pET104/D-TOPO, pET104/D/*lacZ*, pcDNA6/BiotagTM-DEST, pcDNA6/BiotagTM-GW/*lacZ*, pcDNA6/BiotagTM/D-TOPO, pcDNA6/BiotagTM/*lacZ*, pMT/BiotagTM-DEST, and pMT/BiotagTM/GW-*lacZ*.
130. A kit comprising the isolated nucleic acid molecule of claim 1.
131. The kit of claim 130, further comprising one or more components selected from the group consisting of one or more topoisomerases, one or more recombination proteins, one or more vectors, one or more polypeptides having polymerase activity, one or more host cells, and one or more support matrices complexed with avidin or an avidin analog.
132. A kit comprising the isolated nucleic acid molecule of claim 39.
133. The kit of claim 132, further comprising one or more components selected from the group consisting of one or more topoisomerases, one or more recombination proteins, one or more vectors, one or more

polypeptides having polymerase activity, one or more host cells, and one or more support matrices complexed with avidin or an avidin analog.

134. A kit comprising the isolated nucleic acid molecule of claim 80.
135. The kit of claim 134, further comprising one or more components selected from the group consisting of one or more topoisomerases, one or more recombination proteins, one or more vectors, one or more polypeptides having polymerase activity, one or more host cells, and one or more support matrices complexed with avidin or an avidin analog.
136. A host cell comprising a polynucleotide construct that encodes a fusion protein capable of being post-translationally modified, said polynucleotide construct produced according to the method of claim 19.
137. A host cell comprising a polynucleotide construct that encodes a fusion protein capable of being post-translationally modified, said polynucleotide construct produced according to the method of claim 60.
138. A host cell comprising a polynucleotide construct that encodes a fusion protein capable of being post-translationally modified, said polynucleotide construct produced according to the method of claim 104.
139. A method of producing a fusion protein that comprises an amino acid sequence tag, said method comprising:
 - (a) obtaining the host cell of claim 136; and
 - (b) culturing said host cell under conditions wherein said fusion protein is produced by said host cell.
140. The method of claim 139, wherein said amino acid sequence tag is an amino acid sequence that is capable of being post-translationally modified.

141. The method of claim 140, further comprising culturing said host cell under conditions wherein said fusion protein is post-translationally modified in said host cell.
142. The method of claim 140, further comprising culturing said host cell under conditions wherein said fusion protein is biotinylated in said host cell.
143. The method of claim 139, further comprising:
- (a) treating said host cell such that said fusion protein is released from said host cell; and
 - (b) contacting said fusion protein with a detecting composition comprising a molecule that is capable of interacting with said amino acid sequence tag or with a molecular entity that is attached to said amino acid sequence tag.
144. The method of claim 143, wherein said fusion protein is a biotinylated fusion protein, and said detecting composition comprises avidin or an avidin analogue.
145. A method of producing a fusion protein that comprises an amino acid sequence tag, said method comprising:
- (a) obtaining the host cell of claim 137; and
 - (b) culturing said host cell under conditions wherein said fusion protein is produced by said host cell.
146. The method of claim 145, wherein said amino acid sequence tag is an amino acid sequence that is capable of being post-translationally modified.
147. The method of claim 146, further comprising culturing said host cell under conditions wherein said fusion protein is post-translationally modified in said host cell.

148. The method of claim 146, further comprising culturing said host cell under conditions wherein said fusion protein is biotinylated in said host cell.
149. The method of claim 145, further comprising:
- (a) treating said host cell such that said fusion protein is released from said host cell; and
 - (b) contacting said fusion protein with a detecting composition comprising a molecule that is capable of interacting with said amino acid sequence tag or with a molecular entity that is attached to said amino acid sequence tag.
150. The method of claim 149, wherein said fusion protein is a biotinylated fusion protein, and said detecting composition comprises avidin or an avidin analogue.
151. A method of producing a fusion protein that comprises an amino acid sequence tag, said method comprising:
- (a) obtaining the host cell of claim 138; and
 - (b) culturing said host cell under conditions wherein said fusion protein is produced by said host cell.
152. The method of claim 151, wherein said amino acid sequence tag is an amino acid sequence that is capable of being post-translationally modified.
153. The method of claim 152, further comprising culturing said host cell under conditions wherein said fusion protein is post-translationally modified in said host cell.

154. The method of claim 152, further comprising culturing said host cell under conditions wherein said fusion protein is biotinylated in said host cell.
155. The method of claim 151, further comprising:
- (a) treating said host cell such that said fusion protein is released from said host cell; and
 - (b) contacting said fusion protein with a detecting composition comprising a molecule that is capable of interacting with said amino acid sequence tag or with a molecular entity that is attached to said amino acid sequence tag.
156. The method of claim 155, wherein said post-translationally modified fusion protein is a biotinylated fusion protein, and said detecting composition comprises avidin or an avidin analogue.